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TODAY'S Chemist AT WORK

Focus: Process Development

FEATURE

[David Filmore](#)

Tapping chemistry: The brewer's art

Scene: a small bar near the offices of *Today's Chemist at Work*

PHOTOS TAKEN AT OLD DOMINION BREWING CO., ASHBURN, VA

***TCAW* Editor:** *Whew, what a day. Hey barkeep, let's see . . . can I have 473 milliliters of an aqueous solution, heavy on the alpha acid isomers and ester molecules, light on the melanoidins, hold the diacetyl please, and . . .*

Bartender: *Hey, buddy, speak English, willya? We sell beer here, not some kinda chemical formulas.*

***TCAW* Editor:** *Ah, that's where you're wrong, my friend. It's all chemistry—and, well, biology too. Let me explain . . . got a few minutes?*

Bartender: *Uh well, I'm pretty busy . . .*

***TCAW* Editor:** *Okay, then just let me read you the highlights of this article from our magazine. You see, it all begins like this:*

In the modern beer brewing process (for more on its history, see ["Chemistry Chronicles"](#)), four basic ingredients—water, malt, hops (flowers of the *Humulus lupulus* plant), and brewing yeast (*Saccharomyces cerevisiae*)—initiate myriad biological and chemical processes, including enzymatic digestion, fermentation, and isomerization, which must be harnessed to achieve a brew of predictable and consistent quality, whether it's an amber lager, pale ale, a thick porter, or a stout.

Malt-ernatives

An important first step to accomplishing consistent quality is choosing the correct malts. Malts are often made from barley grains, or in some cases, wheat or other grains—and greatly affect the final character of the beer. Before being sent to a brewery, malts are produced in a process in which grain growth is initiated via hydration and germination and then halted by kilning, or heat drying, to preserve the activated enzymes for later use. Kilning at low temperatures yields only lightly colored malts and retains most of the enzymatic activity. Higher-temperature kilning will denature the enzymes to a greater extent and also degrade more of the complex starches and proteins to sugars and amino acids, respectively. At these temperatures, the latter two compounds undergo a condensation reaction, which leads to a sequence of events, including rearrangement, degradation, and polymerization, in a process called the Maillard reaction (common food "browning" chemistry), to form melanoidins—dark, high-molecular-weight compounds with heavy flavors. Therefore, lighter malts are used for pale ale or light pilsner brews, but high-kilned malts are the natural choice for darker beers, such as Munich lagers. Using very high temperatures for kilning produces malts with names such as "caramel", "chocolate", and "black". These have little or no enzymatic power, but are very important flavor and body additives (to supplement an enzymatic malt) for dark and heavy beers, such as porters and stouts.

Once the malts have reached the brewery, the actual purpose of the enzymes comes into play in a step called "mashing". After the malt is ground, it is loaded into a vessel called a "mash tun", where a water suspension is prepared and heated to various temperature levels necessary to overcome the activation barriers of several different enzymes.

Inside of the *lauter tun*—where the wort is separated from the spent grain

Alpha- and beta-amylase are the most important enzymes; they degrade bulky starch molecules into smaller, fermentable sugars (e.g., glucose and maltose) and protease enzymes that break down the complex proteins into soluble material.

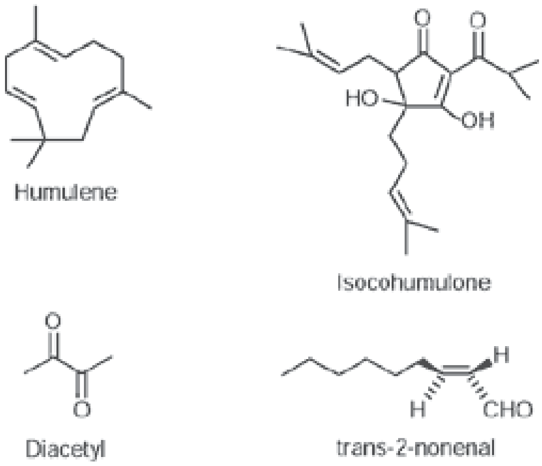
The extent of the enzymatic power will affect how the upcoming fermentation progresses, thus influencing the formation of ethanol and carbon dioxide as well as other important fermentation byproducts. By the same token, the enzyme content will determine the amount of resultant extract that will be digestible by the brewing yeast, and, conversely, the amount of large, unfermentable material, such as dextrins and proteins, that may persist in the final product. Light beers, for example, are generally

produced with high enzyme activity to avoid retention of heavy sugars.

Hop Art
A wide variety of hop cultivars are available, each with a somewhat different array of chemical components and organoleptic properties that offer the potential for an immense palette of different tastes and smells. After mashing, the substance is filtered to create "wort", a clear, sweet solution, which, when boiled, provides a canvas for the hops to work on.

One set of very important hop constituents are α-acids, which isomerize when added to the boiling solution. The resulting α-acid isomers are most responsible for controlling the bitterness of beer, which is a very important aspect, for example, in many English pale ales. Several organic hop oils are available, such as humulene, which derivatize in the boiling process to provide a range of other flavoring and aroma elements—from floral to fruity to spicy—which help define how the beer is perceived overall.

For bittering purposes, hops are usually added early in the boil, so the isomerizations can go to completion. Alternatively, in some cases, pre-isomerized hop extracts are added later to achieve similar results. For less bitter beers (many lagers fall into this category), hops are predominantly included late in the boil or even after the solution is cooled. This allows volatile oil compounds to survive in the product and prevents extensive acid isomerization.



Good and Bad of Beer. The structures of four compounds that may be on the brewer's mind: Humulene is a flavor compound from hop oils, and isocohumulone is an isomerized hop alpha acid. Diacetyl, from fermentation, and *trans*-2-nonenal, from oxidation, are both known to have unstable flavor characteristics.

Large numbers of iso-α-acids and hop oil derivatives have been isolated from beer by chemists, but this has not led to a clear correlation between constituents and overall hop character. It seems that the final product depends largely on complex, and not well understood, synergistic effects between the species; thus, successful hop addition depends much on experimentation and experience.

Yeastly Decisions
As already suggested, one of the main functions of wort constituents is to provide nutrients that will fuel the fermentation by brewing yeast. Amino acids and peptides (formed in the mash via proteases) provide essential

nitrogen sources for yeast growth. Fermentable sugars are broken down into glucose, which is transformed into the main products ethanol and CO_2 through a progression of enzymatic processes. In addition, the major intermediate in the fermentation process, pyruvic acid, and its final product, ethanol, both undergo side reactions to form smaller amounts of some organic products, such as ketones, esters, and higher alcohols, which can strongly influence flavor. The quantity and ratios of different sugars, amino acids, and peptides will significantly affect the extent to which each product is produced.

Other than wort content, the choice of brewing yeast—always a strain of *Saccharomyces cerevisiae*—is a major factor in fermentation. Varying energy sources and enzymes in strains of *S. cerevisiae* leads to differences in fermentation effectiveness, duration, and conditions. Yeast strains vary widely, so microbiological analysis must be carried out to determine how the yeast and the particular wort will interact. Often, brewing yeast is put into one to two categories—"top-fermenting" ale yeast, so-called because of its common property of flocculating to the top of the tank following fermentation, and "bottom-fermenting" lager yeast. Ale yeast strains tend to work best at temperatures between 10–25 °C over about 40 h, which tends to favor more significant formation of organic byproducts, such as esters, which are known for their fruity fragrances, the central aroma components for many types of ale. Lager strains are usually used at lower temperatures (5–15 °C) for longer periods of time and produce less byproduct, yielding the less-pronounced flavors characteristic of lagers.

Monitoring the Mix

To brew a consistent product, industrial brewers require detailed analysis throughout the production process from chemists, microbiologists, and other scientists. According to James Munroe, director of Brewing Technical Services at Anheuser-Busch, "all incoming materials are inspected and tested to meet quality specifications." Testing methods include sensory evaluations, such as tasting and smelling hops; physical assessments, such as measuring malt moisture; and chemical analyses, including malt enzyme testing.

Cylindroconical fermentation tanks

The stages of production that require laboratory testing, says Munroe, "are finished wort prior to yeast addition, end of fermentation beer, end of lagering, filtered beer, and packaged beer." A range of analytes are investigated, including ethanol, total free amino nitrogen, and microorganisms (including brewing yeast, which is generally recovered and reused, and potential contaminants). Gas chromatography is commonly used to measure some of beer's volatile flavor components, and HPLC is

of growing importance in brewery analytical labs for analyzing carbohydrates and some hop compounds. Traditional microbiological plates are also regular elements of brewery testing, although newer techniques, such as DNA microarrays, are becoming more popular. Additionally, measurements of such parameters as pH, solids content, and specific gravity are vital elements to controlling a beer's properties. To improve the time between monitoring and adjustment, says Munroe, "the current trend is to introduce in-line instruments for characteristics that can be used for process control, such as pH, solids, turbidity, dissolved O_2, and ethanol."

Cleaning Up

This limited discussion reveals the ongoing complexities of the brewing process. Inevitably, unwanted products are generated, so several built-in purification steps are part of the process. For example, filtration extraction is carried out

- to remove unmodified grain components, following the mash;
- to extract protein–polyphenol complexes (polyphenols are a hop constituent), which can cause haze and sedimentation, after the hopped wort is cooled; and
- in the final (preceding carbonation and packaging) "cold filtering" process after the beer has been matured and conditioned for a period of weeks to months.

Chemical steps are also taken to remove unwelcome components such as some diketones, particularly diacetyl (2,3-butadione), which is a fermentation byproduct with an unstable flavor profile. After primary fermentation, temperatures are often raised ("diacetyl rest") to allow the yeast enzymes to further reduce these species to less flavor-active diols. Furthermore, during the subsequent conditioning step, the beer commonly undergoes a secondary fermentation at low temperatures to remove all lingering fermentable extract. And to provide more clarity to the beer's appearance, additional agents, such as proteolytic enzymes or silica gel, are added during the conditioning step to remove lingering protein–polyphenol complexes.

Some flavor-fouling components should be prevented from showing up in the first place. "The only time oxygen is permitted in the process," says Ray Klimovitz, technical director of the Master Brewers Association of

All in the Head

Bubbles journeying upward and resting on their frothy bed—a salient image of beer. It comes to be by the interaction of carbon dioxide with complexes made of large, hydrophobic barley proteins that have survived brewing along with lipids, iso-α-acids, and polyphenols. As the CO_2 rises, it is coated by these complexes, which, if substantial enough, will preclude, at

the Americas, “is for the aeration of the yeast immediately at the start of fermentation. Otherwise it is taboo!” Oxidation of malt lipids to flavor-unstable aldehydes is a particular problem. *Trans*-2-nonenal is one common example; it leads to a stale, cardboard-like flavor.

Therefore, oxygen is minimized via various process engineering strategies, such as deaerating water, using inert gas atmospheres in brewing vessels, and avoiding high levels of oxygen-attracting copper in the equipment. In addition, substances with antioxidant properties can be added at various stages.

least temporarily, the bubbles from bursting.

The extent to which a head will form and persist largely depends on the survival of the foam-forming barley proteins through the malting and brewing process. However, conditions that favor retention may also cause slightly smaller protein complexes, which may be haze-forming, to hang around—adding up to a very delicate balance for brewers to deal with.

Researchers looking to provide a greater understanding of how foam can be successfully controlled are studying the chemical pathways of these head-forming proteins throughout the brewing process (see *J. Agric. Food Chem.* **2000,** *48*, 5023–5029). In addition, analytical techniques are being developed to monitor these species. For instance, UK researchers have developed a monoclonal antibody probe for hydrophobic beer proteins, which could have use as an in-process quality control method for foam stability (*J. Agric. Food Chem.* **1999,** *47*, 3044–3049).

Of course, another approach, which, notably, is taken by the brewers of Guinness stout, is to add nitrogen, which forms bubbles that are more stable and less likely to burst.

Brewing Beyond

Beer brewing is often said to be a mixture of science and art. We’ve covered some of the science, but there are many aspects of the brewing process that remain elusive, and the roles of each of the hundreds of compounds that make their way into the final brew are not clear. Therefore, brewers must rely on experience (and some creativity) to make decisions; however, researchers are working on unearthing some of the subtleties of the science of brewing to help take some of the guesswork out of the process.

For example, detailed mechanistic studies of the formation and degradation of enzymes during malting are being carried out. Researchers are also using sophisticated GC-olfactometry methods to explicitly match hop-derived compounds with flavor and aroma components. Yeast, as one might guess, is also a primary subject of beer-related research, and delving into genomics has led scientists to experiment with genetically modified yeast

strains to accomplish such feats as the ability to use larger, normally unfermentable sugars or to chemically bypass the formation of diacetyl. There are a host of other research projects "brewing" on the topic. Organizations such as the American Society of Brewing Chemists (ASBC) provide a centralized medium for industry and academic research in their journal, annual meetings, and various other tools. Just perusing the abstracts of the *Journal of the ASBC* and other journals, including ACS's *Journal of Agriculture and Food Chemistry,* one can get a feel for the many issues being investigated—all in the name of providing more control over the process and a clearer picture of what makes up the final product.

Bartender (yawning): *Alright already, I think I get the idea.*

***TCAW* Editor:** *Well then, my work here is finished. Pass me a pint of good old brewski and we'll toast the holidays.*

Further Reading

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Charlie Bamforth, Professor of Brewing Science, University of California–Davis, Web site.
http://foodscience.ucdavis.edu/bamforth/index.html.

David Filmore *is an assistant editor of* Today's Chemist at Work. *Send your comments or questions regarding this article to* tcaw@acs.org *or the Editorial Office, 1155 16th St N.W., Washington, DC 20036.*

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